CLAIMS

1. A method which is prognostic for a preneoplastic/neoplastic disease afflicting a subject vertebrate, wherein said disease affects a tissue, which tissue normally expresses MN/CA IX protein, but loses or has significantly reduced MN/CA IX expression upon carcinogenesis, said method comprising:

(a) detecting MN/CA9 gene expression product in a sample comprising preneoplastic/neoplastic tissue taken from said vertebrate,

(b) quantitating the level of said MN/CA9 gene expression product in said sample,

(c) comparing the level of MN/CA9 gene expression product of step (b) to the average level of MN/CA9 gene expression product in comparable samples taken from vertebrates afflicted by the same preneoplastic/neoplastic disease as the subject vertebrate, and

(d) determining that said subject vertebrate has a poorer prognosis if the level of MN/CA9 gene expression product of step (b) is higher than the average level of MN/CA9 gene expression product in said comparable samples;

wherein said MN/CA IX protein is encoded by a nucleotide sequence selected from the group consisting of:

(3) SEQ ID NO: 1's coding region;

(2) nucleotide sequences that hybridize under stringent hybridization conditions of 50% formamide at 42 degree C. to complement of SEQ ID NO: 1's coding region; and

(3) nucleotide sequences that differ from SEQ ID NO: 1's coding region or from the nucleotide sequences of (2) in codon sequence due to the degeneracy of the genetic code.

2. The method of Claim 1 wherein 40% or more of the cells of said tissue, when unaffected by said preneoplastic/neoplastic disease, express MN/CA IX protein.

3. The method of claim 1, wherein said preneoplastic/neoplastic disease afflicting said subject vertebrate is selected from the group consisting of preneoplastic/neoplastic diseases of gastric mucosa, gallbladder, biliary ducts, ductal

cells of duodenal glands, testis including ductular efferens and rete testis, ovary including surface coelomic epithelium and rete ovari, basal cells of hair follicles, and central nervous system choroid plexus.

4. The method of claim 1 wherein said vertebrate is a mammal, and wherein said preneoplastic/neoplastic disease is selected from the group consisting of preneoplastic/neoplastic diseases of gastric mucosa, gallbladder, bilary ducts, and ductal cells of duodenal glands.

5. The method of claim 1 wherein said vertebrate is a human patient, and said preneoplastic/neoplastic disease is selected from the group consisting of neoplastic diseases of gastric mucosa, gallbladder, biliary ducts and ductal cells of duodenal glands.

6. The method of claim 1 wherein said neoplastic disease is gastric cancer, and wherein said sample is taken from the invasion front of said gastric cancer.

7. The method of claim 5 wherein said neoplastic disease is a tumor, and said sample is taken from said tumor and/or from a metastatic lesion derived from said tumor.

8. The method of claim 1, wherein immunohistochemical staining with MN/CA IX-specific antibody is used to detect and quantitate MN/CA IX protein in the sample, and wherein the quantitating step (b) comprises determining an immunoreactivity score of cells in said sample comprising:

(b1) determining the percentage of immunoreactive cells, wherein the percentage of immunoreactive cells is assigned

a value of 0 if no immunoreactive cells,

a value of 1 if less than 10% immunoreactive cells,

a value of 2 if between 11% and 50% immunoreactive cells, or

a value of 3 if more than 50% immunoreactive cells;

(b2) determining the intensity of immunostaining of the immunoreactive cells, wherein the intensity of MN/CA IX immunostaining is assigned

a value of 0 for staining equal to a negative control,

a value of 1 for weak staining,

a value of 2 for moderate staining, or

a value of 3 for strong staining; and

(b3) adding the value for the percentage of immunoreactive cells found in step (b1) and the value for the intensity of immunostaining found in step (b2) to obtain the immunoreactivity score;

wherein the comparing step (c) comprises determining the immunoreactivity scores of said comparable samples analogously to the determination of the immunoreactivity score of the sample from the subject vertebrate in steps b(1) to b(3), and averaging said immunoreactivity scores from said comparable samples; and

wherein if the immunoreactivity score of the sample determined in steps b(1) to b(3) is above the average immunoreactivity score of said comparable samples, concluding in step (d) that said vertebrate has a poorer prognosis than if said immunoreactivity score is at or below said average immunoreactivity score.

9. The method of claim 1, wherein a poorer prognosis is measured in terms of shortened survival, increased risk of recurrence of said preneoplastic/neoplastic disease, or in diminished or refractory response to treatment.

10. The method of claim 1, wherein said disease is neoplastic and comprises a tumor, or a tumor and one or more metastatic lesions derived from the tumor, and wherein a poorer prognosis is measured in terms of shortened survival, increased risk of recurrence of said neoplastic disease, or diminished or refractory response to treatment, following treatment and/or surgical removal of the tumor, or the tumor and said one or more metastatic lesions.

11. The method of claim 1, wherein said preneoplastic/neoplastic sample is a formalin-fixed, paraffin-embedded tissue sample or a frozen tissue sample.

12. The method of claim 1, wherein said MN/CA9 gene expression product comprises an MN/CA IX protein or MN/CA IX polypeptide.

13. The method of claim 1, wherein said MN/CA9 gene expression product comprises an mRNA encoding an MN/CA IX protein or MN/CA IX polypeptide or a cDNA encoding an MN/CA IX protein or MN/CA IX polypeptide.

14. The method according to claim 1, wherein said detecting step (a) comprises the use of an assay selected from the group consisting of Western blots, enzyme-inked immunosorbent assays, radioimmunoassays, competition immunoassays, dual antibody sandwich assays, immunohistochemical staining assays, agglutination assays, fluorescent immunoassays and cytofluorometry.

15. The method of claim 1, wherein said detecting step (a) is by PCR, RT-PCR, real-time PCR, or by quantitative real-time RT-PCR.

16. The method according to claim 1, wherein said detecting step (a) comprises the use of the monoclonal antibody secreted by the hybridoma VU-M75 which has Accession No. ATCC HB 11128.

17. The method according to claim 1, wherein said detecting step (a) is by immunohistochemical staining, and wherein said quantitating step (b) comprises determining the percentage of immunoreactive cells.

18. The method according to claim 1, wherein the detecting step (a) is by immunohistochemical staining, and wherein the quantitating step (b) comprises determining the percentage and the intensity of immunostaining of immunoreactive cells.

19. The method of claim 1, wherein said vertebrate is a mammal.

20. The method of claim 19, wherein said mammal is a human.

21. The method of claim 1, wherein said prognostic method is used as an aid in the selection of treatment for said preneoplastic/neoplastic disease afflicting said vertebrate.

22. The method of claim 1 wherein said sample is taken from the invasion front of said preneoplastic/neoplastic disease, and said comparable samples are analogous invasion front samples.

23. The method of claim 22 wherein said preneoplastic/neoplastic disease is a neoplastic disease.

24. A method which is prognostic for a preneoplastic/neoplastic disease afflicting a subject vertebrate, wherein said disease affects a tissue in which 40% or more of the cells normally express MN/CA IX protein, but said tissue loses or expresses MN/CA IX at a significantly reduced level upon carcinogenesis, said method comprising:

(a) taking a tissue sample from the invasion front of said preneoplastic/neoplastic disease;

(b) detecting in said invasion front sample whether MN/CA9 gene expression product is absent or at a significantly reduced level from the level that said MN/CA9 gene expression product is normally expressed in said tissue, when said tissue is unaffected by said disease; and

(c) concluding that if said MN/CA9 gene expression product is neither absent nor at such a significantly reduced level in said invasion front sample, that the subject vertebrate has a poorer prognosis than if said MN/CA9 gene expression product were absent or at a such a significantly reduced level in said invasion front sample.